

Appl. No. 10/291,342
Amdt. dated 07/22/2005
Reply to Office action of 06/16/2005

AMENDMENTS TO THE CLAIMS

In the claims, please cancel claim 8 and amend claims 1, 7, and 9-14 as follows:

1. (currently amended) A process for enhancing transposase mediated integration of a transposon into a target nucleic acid in a cell-free system comprising:
 - a) forming an integrator complex;
 - b) combining said integrator complex with an ~~enhancing~~ transfection reagent; and,
 - c) combining said integrator complex and said ~~enhancer~~ transfection reagent with the target nucleic acid in a cell-free system.
2. (previously presented) The process of claim 1 wherein the transposase comprises Tn5 transposase.
3. (previously presented) The process of claim 2 wherein the Tn5 transposase comprises hyperactive Tn5 transposase.
4. (previously presented) The process of claim 3 wherein the hyperactive Tn5 transposase comprises the EK54MA56LP372 mutant Tn5 transposase (SEQ ID 5).
5. (previously presented) The process of claim 1 wherein the transposon comprises a Tn5 transposon.
6. (previously presented) The process of claim 5 wherein the Tn5 transposon is flanked by elements selected from the group consisting of Tn5 outer elements, Tn5 inner elements, and Tn5 mosaic elements.
7. (currently amended) The process of claim 1 wherein the ~~enhancer~~ transfection reagent is selected from the group consisting of polycations, cationic polymers and cationic lipids.
8. (canceled)
9. (currently amended) The process of claim ~~[[7]] 1~~ wherein the ~~cationic polymer~~ transfection reagent consists of polyethyleneimine.
10. (currently amended) The process of claim ~~[[7]] 1~~ wherein the ~~enhancing~~ transfection reagent consists of both cationic proteins and cationic lipids.

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11. (currently amended) The process of claim ~~[[8]]~~ 1 wherein the transfection reagent is selected from the group consisting of TRANSIT[®]-LT1, TRANSIT[®]-LT2, TRANSIT[®]-INSECTA, TRANSIT[®]-HELAMONSTER[™], TRANSIT[®]-100, TRANSIT[®]-293, TRANSIT[®]-KERATINOCYTE, LIPOFETIN[®] and TRANSIT[®]-EXPRESS.
12. (currently amended) The process of claim ~~[[7]]~~ 1 wherein the ~~cationic polymers are~~ transfection reagent is selected from the group consisting of poly-L-lysine (PLL) and poly-D-lysine.
13. (currently amended) A process for integrating a nucleic acid into a target nucleic acid comprising making a transposon, associating a transposase with the transposon to form an integrator complex, combining the integrator complex and a cationic ~~enhancing~~ transfection reagent together in solution, and incubating the composition with a target nucleic acid in a cell-free system, wherein the transposase integrates the transposon into the target nucleic acid.
14. (currently amended) A kit for cell-free integration of a nucleic acid into a target nucleic acid comprising a receptacle containing a transposase and a receptacle containing an ~~enhancing a transfection reagent wherein the enhancing reagent is selected from the group consisting of transfection reagents, polycations, cationic polymers and cationic lipids.~~

AMENDMENTS TO THE CLAIMS

Please cancel claims 18-20 and amend claim 1 as follows:

1. (currently amended) A process for *in vivo* expression of longer than seven days of a non-viral, linear DNA nucleic acid sequence from a delivered expression cassette, comprising:
 - a) providing the expression cassette comprising the nucleic acid sequence operably linked to a promoter;
 - b) forming a non-viral, linearized plasmid DNA vector comprising the expression cassette; and,
 - c) delivering the non-viral, linearized plasmid DNA vector to a hepatocyte in a mammal, wherein providing the expression cassette on the non-viral, linearized plasmid DNA vector results in increased expression in the hepatocyte after seven days defined by at least 20% more gene product than is expressed from a supercoiled plasmid from which the linearized plasmid is derived of longer than seven days of the nucleic acid sequence.
2. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains blunt ends.
3. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains sticky ends.
4. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains a blunt end and a sticky end.
5. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector is generated by restriction enzyme digestion.
6. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector is generated by polymerase chain reaction.
7. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains an expression cassette isolated from a plasmid backbone.

8. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains an expression cassette which is flanked by sequence derived from inner Tn5 transposase recognition elements.
9. (previously presented) The process of claim 8, wherein the non-viral, linear DNA vector ends are blunt.
10. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains an expression cassette which is flanked by sequence derived from inner outer Tn5 transposase recognition elements.
11. (previously presented) The process of claim 10, wherein the non-viral, linear DNA vector ends are blunt.
12. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector contains an expression cassette which is flanked by chimeric ends derived from Tn5 transposase recognition elements.
13. (previously presented) The process of claim 12, wherein the non-viral, linear DNA vector ends are blunt.
14. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector is delivered to cells intravascularly.
15. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector are delivered intravascularly using pressure.
16. (canceled)
17. (previously presented) The process of claim 1, wherein the non-viral, linear DNA vector is delivered by direct interstitial injection.
18. (canceled)
19. (canceled)
20. (canceled)